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Comparison of Trihalomethanes in Tap Water and Blood

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Trihalomethane (THM) concentrations in blood and tap water were measured for 50 women living in two locations with different bromide concentrations and disinfectant types. Blood samples were taken from each woman early in the morning prior to any major water-use activity and again immediately after showering. Each residence was sampled for THMs in tap water prior to the woman's shower. Cobb County, GA, tap water exhibited high THM concentrations composed primarily of chloroform. Corpus Christi, TX, tap water exhibited lower THM concentrations with significant proportions of brominated THMs. THMs in tap water and blood were compared using mole fraction speciation, extent of bromine incorporation, and correlation analysis. Results indicated that THMs in the blood rose significantly as a result of showering, that showering shifted the THM distribution in the blood toward that found in the corresponding tap water, and that THMs measured in the blood of women living in the two locations reflected species and concentration differences in their respective tap waters. In general, blood concentrations were not significantly correlated with tap water concentrations. This finding suggests that other factors, in addition to tap water concentrations, may be important in determining THM concentrations in the blood.

Introduction

Trihalomethanes (THMs) are a class of disinfection byproducts formed when chlorine reacts with natural organic matter and bromide found in drinking water. They are comprised of four compounds: chloroform ($CHCl_3$), bromodichloromethane ($CHCl_2Br$), dibromochloromethane ($CHClBr_2$), and bromoform ($CHBr_3$). THMs became a public health concern due to their suspected carcinogenic nature (*1*, *2*). In November 1998, the United States Environmental Protection Agency promulgated the Stage 1 Disinfectants/Disinfection By-Products Rule that lowered the maximum contaminant level of the four THM species (THM4) in finished drinking water from 100 to 80 μg/L. These standards are based on a running annual average calculated from quarterly THM4 measurements (*3*).

Recent epidemiological studies have shown that exposure to THMs in tap water may be associated with adverse reproductive outcomes such as spontaneous abortion, birth defects, prematurity, low birth weight, and intrauterine growth retardation (*4*–*9*). However, the studies proposing these relationships were limited by a lack of reliable exposure characterization (*10*). Epidemiological studies that examine adverse reproductive outcomes use exposure windows of weeks to several months (*6*, *8*–*11*). Unfortunately, the availability of THM data in water distribution systems with sufficient spatial and temporal resolution to assess these types of acute health outcomes is limited. Water utilities are required to measure THMs on a quarterly basis from four sampling locations. A further limitation of the epidemiological studies referenced above is that most considered only ingestion, whereas methods of assessing THM exposure in women of reproductive age are needed that consider all potential routes of exposure. THMs are ingested during consumption of hot and cold chlorinated tap water, but because THMs are volatile, they may also be inhaled during water-use activities such as showering, bathing, dishwashing, or clothes washing. Showering, bathing, and other activities that involve contact between tap water and skin can also lead to THM exposure via dermal absorption.

Several indicators of THM exposure have been proposed, including measurement of THM concentrations in drinking water or ambient air (*12*, *13*) and use of biomarkers. Various biological samples (including alveolar air, blood, mother's milk, and adipose tissue) have been used to measure internal dose levels of THMs (*14*). Studies have shown that blood and breath concentrations respond to exposure in a very similar manner (*15*). Although breath samples are less invasive and lead to better subject participation, blood levels are generally more sensitive to low exposures. In most cases, THM concentrations are below detection limit in breath before an exposure occurs (*16*). By contrast, blood levels of the chlorinated THMs have been measured at detectable levels before significant exposure (*17*). Blood consists of both organic (lipid) and lipophobic regions. A significant amount of the THMs will partition into the lipid phase of blood and fat tissue, but there will be an equilibrium that allows the quantitative determination of THMs in blood. Since internal dose levels both prior to and after exposure were of interest in this study, blood was selected for biological sampling. Blood concentrations are also thought to represent an integrative measure of all routes and sources of exposure (*18*).

As chlorinated tap water is thought to be the primary source of THMs (*19*), internal dose levels in the blood should be related to tap water concentrations and water-use activities. Accordingly, the goal of this work was to explore tap water concentrations as an indicator of THM exposure by comparing concentrations in participants' tap water to concentrations measured in their blood.

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Methods

During the summer of 1999, a total of 50 study participants were evaluated from two geographic areas. Participants were selected from mothers who resided in Cobb County, GA, or Corpus Christi, TX. The water utility serving Cobb County, GA, uses free chlorine as a primary and secondary disinfectant, and bromide levels in the source water are relatively low with concentrations of 30 μg/L or less. THM levels are relatively high with the distribution heavily favoring the more chlorinated species (i.e., chloroform). The water utility serving Corpus Christi (Nueces County), TX, uses combined chlorine for primary and secondary disinfection, and the bromide levels in the source water are relatively high with typical concentrations on the order of 400 μg/L. THM levels in the water are lower than typically measured in systems using free chlorine, and the distribution of THMs favors the brominated species. The study was conducted over a 4-week period in each study location. Each of the 25 participants in each study location was visited and sampled once over this 4-week period. Each woman was asked to record her water-use activities in a diary for 36 h, and water flows were recorded for 24 h using a data logger attached to the water meter. The selection procedures and components of the study not directly relevant to this paper have been described in detail elsewhere (*20*).

A member of the study team collected duplicate tap water samples from the kitchen faucet (or nearest unfiltered tap) in each home early in the morning at the time the participant showered. Faucets were set to the "coldest" position, but water temperatures were not measured at the tap. Samples were collected headspace-free in 40-mL acid-washed vials after letting the faucet run for 5 min. The chlorine residual was quenched using ammonium sulfate (Mallinckrodt, Phillipsburg, NJ) to prevent further THM formation. After collection, samples were refrigerated and on a weekly basis were packed into coolers with ice packs and sent by overnight delivery to the University of North Carolina for analysis. Samples were held in a refrigerator at 4 °C until analysis, which was typically completed during the following week. The THMs were extracted using high-purity pentane (Burdick & Jackson, Muskegon, MI) and were analyzed on a Hewlett-Packard (Avondale, PA) 5890 series II gas chromatograph using electron capture detection based on Standard Method 6232 (*21*). The separation was accomplished using a J&W Scientific (Folsom, CA) DB-1 capillary column. 1,2-Dibromopropane (Aldrich Chemical Co., Milwaukee, WI) was used as an internal standard during the extraction process; checkpoints and spiked samples were used for quality assurance during GC analysis. Calibration curves were developed for each set of samples using regression analysis of relative response factors compared with standard concentrations. The THM concentrations reported are the averages of the duplicate samples.

A blood sample was taken from the participant immediately before showering and, in most cases, prior to any major water-use activity to represent a "baseline" exposure level. A blood sample was also taken as soon after showering as was practical. The purpose of this sample was to estimate "peak" blood THM levels, although it is recognized that THM levels are expected to be at their highest at the end of the shower. Collection of samples closer to the termination of the shower was not always practical. Blood samples were collected in "gray-top" Vacutainer tubes that were specifically processed to remove contamination from volatile organic compounds (VOCs) (*22*), shipped by overnight carrier to the Centers for Disease Control and Prevention in Atlanta, and stored at 4 °C until analysis. The blood samples were analyzed for THMs using a purge-and-trap/gas chromatography/isotope-dilution mass spectrometry method with detection limits in the nanograms per liter range. Extraction of VOCs from blood, trapping on Tenax in a glass-lined steel tube, removal of water, and concentration on a liquid nitrogen trap were done with a Tekmar (Cincinnati, OH) 3000 purge-and-trap concentrator with an attached ALS 2016 automated sampler. The analytes were separated with a J&W Scientific (Folsom, CA) DB-624 column mounted in a Hewlett-Packard (Avondale, PA) model 5890 series II gas chromatograph. Mass spectral analysis was done with a Micromass (Beverly, MA) Ultima high-resolution mass spectrometer that operated at 10 000 resolving power (5% valley definition) in the SIR-voltage mode. Masses were calibrated against a mixture of low-boiling perfluorokerosene (GB Scientific, Novato, CA) with a 50/50 mixture of toluene and benzene. Peak areas were determined using the OPUSquan software of Micromass. Blood concentrations were determined by regression analysis of relative response factors against compound weight.

TABLE 1. Minimum Detection Limits (MDLs) and Maximum Reporting Levels (MRLs) for THMs in Blood and Tap Water

	blood		water	
THM species	MDL (ng/L)	MRL (ng/L)	MDL (μg/L)	MRL (μg/L)
$CHCl_3$	2.5	434	2.5	125
$CHCl_2Br$	0.21	93	1	100
$CHClBr_2$	0.24	93	1	100
$CHBr_3$	0.23	96	1	100

The THMs in blood and tap water were compared using the THM speciation based on mole fractions, the extent of bromine incorporation using the bromine incorporation factor (BIF; see below), and the correlations between THM concentrations measured in corresponding blood and tap water samples. THM concentrations that were observed to be below detection levels were assigned one-half that value for data analysis (see Table 1). The minimum detection level (MDL) was determined as the lowest concentration that elicited a response by the GC-ECD. The maximum reporting level (MRL) was the highest concentration used to develop the calibration curves. If the calibration curve exhibited nonlinear behavior at high concentrations, the MRL was set as the highest concentration that exhibited linear behavior. THM concentrations that were calculated to be beyond the range of the calibration curve were reported as greater than the MRL of the calibration curve for analysis purposes (i.e., >MRL). Mole fractions (see eq 1) were used for some analyses to normalize the individual THM species concentrations and to compare proportions of individual species rather than absolute concentrations. In eq 1, x_i represents the mole fraction of species i, C_i represents the molar concentration of species i, and C_{THM4} represents the total molar concentration of all four THM species:

$$x_i = \frac{C_i}{C_{THM4}} \quad (1)$$

Frequency distributions for occurrence of the individual THM species were evaluated for both tap water and blood samples. For each THM species, location (Cobb County or Corpus Christi), sample type (tap water or blood), and frequency distributions were developed using a mole fraction interval of 0.1 as the bin size.

The bromine incorporation factor (BIF) represents the average number of moles of bromine per mole of THM4 (*23*). The BIF was calculated by dividing the sum of the molar concentration of bromine in each individual THM species by the total molar concentration of THM4, as shown by eq 2. Each term in the numerator of eq 2 is a molar concentration of an individual THM species multiplied by the number of

TABLE 2. Range of THM4 and Individual Species Concentrations (μg/L) Measured in Tap Water and in Before Showering (BS) and After Showering (AS) Blood Samples

		Cobb County, GA			Corpus Christi, TX		
concn (μg/L)		tap water ($n = 24$)	blood[a] (BS) ($n = 24$)	blood[a] (AS) ($n = 24$)	tap water ($n = 24$)	blood[a] (BS) ($n = 24$)	blood[a] (AS) ($n = 24$)
THM4	maximum	131	0.176	>0.543	59	>0.507	>0.658
	minimum	52	0.044	0.155	16	0.014	0.083
	median	100	0.080	0.313	44	0.044	0.172
$CHCl_3$	maximum	112	0.170	>0.430	15	>0.430	>0.430
	minimum	42	0.037	0.130	2	0.009	0.025
	median	85	0.070	0.280	8	0.025	0.057
$CHCl_2Br$	maximum	17	0.017	0.093	15	0.035	0.083
	minimum	5	0.002	0.017	5	0.002	0.009
	median	14	0.006	0.038	12	0.007	0.046
$CHClBr_2$	maximum	4	0.003	0.029	20	0.031	>0.093
	minimum	<1	0.001	0.003	5	0.002	0.011
	median	2	0.001	0.006	14	0.007	0.042
$CHBr_3$	maximum	<1	0.0052	0.0059	17	0.021	0.064
	minimum	<1	0.0001	0.0001	2	0.001	0.006
	median	<1	0.0003	0.0005	9	0.004	0.018

[a] THM4 values reported as greater than some value indicate that the concentration of one of the species was outside the linear range of its calibration curve. In those instances, the individual THM species was assigned its maximum level and summed with the concentrations of the other three THM species to obtain a THM4 value.

bromine atoms in the species, and C_{THM4} represents the total molar concentration of all four THM species. The BIF can be used to quantitatively describe the degree of bromination in the THM distribution:

$$BIF = \frac{C_{CHCl_2Br} + 2C_{CHClBr_2} + 3C_{CHBr_3}}{C_{THM4}} \quad (2)$$

BIF values, calculated for each location and sample type, range from 0.0 if chloroform is the only THM species present to 3.0 if bromoform is the only THM species present. Frequency distributions were developed for BIF values using an interval size of 0.2.

Statistical tests were conducted using Stata version 6.0 software (Stata Corporation, College Station, TX). Distributions of the THM4 and individual species were tested for normality using the Shapiro–Wilk statistic, which is appropriate for the sample sizes found in this study (*24*, *25*). Nonparametric tests were then employed for two reasons: (i) almost all of the species concentrations were determined to be nonnormally distributed and (ii) arithmetic means could not be calculated accurately due to the existence of >MRL values. THM4 concentrations in the blood before and after showering were compared within each study group (Cobb County or Corpus Christi) using a Wilcoxon signed-rank test (*26*) to determine if blood concentrations significantly rose as a result of that activity. Blood samples taken before showering were compared between the study groups using Wilcoxon rank sum tests (*26*) to determine if "baseline" exposures between the two groups were significantly different. Within each study group, the distributions of THMs in tap water, blood before showering, and blood after showering were compared using Kruskal–Wallis tests (*26*) to examine how showering affected blood THM speciation. Kruskal–Wallis tests were also used to compare BIF distributions in the tap water, blood before showering, and blood after showering.

Blood concentrations were analyzed against tap water concentrations to determine the correlation, if any, between the two. Spearman's correlation coefficients (*26*), denoted as r_s, were used to quantify the correlations for overall THM4 concentrations and individual THM species within each study group. Significance (p) values were reported up to the 10% level due to the small sample size.

Results

Cobb County, GA. THM4 concentrations measured in Cobb County tap water in the homes of participants were relatively high and strongly favored the chlorinated species. The range of THM4 concentrations was 52–131 μg/L, with a median THM4 concentration of 100 μg/L (see Table 2). (These THM4 values were measured under drought conditions in August 1999 and are not representative of typical distribution system values in Cobb County. The water utility's running annual average for THM4 at the time of the study was 57 μg/L.) The predominant THM species was consistently chloroform, with a median concentration of 85 μg/L. Bromoform was not detected in any of the tap water samples.

Blood samples taken from Cobb County participants prior to showering contained THM4 concentrations ranging from 0.044 to 0.176 μg/L, with a median concentration of 0.080 μg/L (see Table 2). These values are approximately 3 orders of magnitude lower than those measured in the tap water. Blood samples taken after showering showed that blood THM levels increased significantly as a result of that water-use activity ($p < 0.0001$). The median THM4 concentration measured in the blood after showering was 0.313 μg/L, with a range from 0.155 to >0.543 μg/L. This represents a 4-fold increase in the median THM4 concentration measured in the blood. Nine of the 24 subjects had chloroform concentrations after showering that exceeded the MRL.

An analysis of the distribution of THM species revealed that chloroform was the predominant THM species in the tap water samples, as well as in both sets of blood samples. The median chloroform mole fraction measured in the tap water, before-showering (BS) blood samples, and after-showering (AS) blood samples was 0.880, 0.919, and 0.890, respectively (see Table 3). These differences in chloroform mole fractions were found to be statistically significant ($p = 0.0001$), which indicates that showering caused the proportion of chloroform in the blood to decrease toward that found in the tap water.

BIF values were calculated to quantitatively compare the degree of bromination among the three sets of samples. The median BIF for tap water, BS blood samples, and AS blood samples were 0.138, 0.090, and 0.123, respectively (see Table 3). Although each set of samples averaged approximately 1 mol of bromine/10 mol of THM4, the three BIF distributions were found to be statistically different ($p = 0.0001$). Showering

TABLE 3. Median Mole Fractions for THM Species in Tap Water and Before Showering (BS) and After Showering (AS) Blood Samples As Well as Median Bromine Incorporation Factors (BIFs)

	Cobb County, GA			Corpus Christi, TX		
THM species or BIF	tap water ($n = 24$)	blood (BS) ($n = 24$)	blood (AS) ($n = 24$)	tap water ($n = 24$)	blood (BS) ($n = 24$)	Blood (AS) ($n = 24$)
$CHCl_3$	0.880	0.919	0.893	0.286	0.680	0.427
$CHCl_2Br$	0.106	0.072	0.094	0.287	0.128	0.260
$CHClBr_2$	0.011	0.009	0.013	0.275	0.114	0.210
$CHBr_3$	0.002	0.002	<0.001	0.148	0.045	0.065
BIF	0.138	0.090	0.121	1.285	0.549	0.910

caused the distribution of BIF values to increase toward that found in the tap water.

Corpus Christi, TX. THM4 levels measured in Corpus Christi tap water were lower than in Cobb County, ranging from 16 to 59 μg/L, with a median concentration of 44 μg/L (see Table 2). All four THM species were measured at detectable levels. Dibromochloromethane was the predominant species on a mass basis with a median concentration of 14 μg/L, although chloroform and bromodichloromethane had comparable molar concentrations due to the difference in molecular weights among the compounds. Bromoform was measured at concentrations ranging from 2 to 17 μg/L.

The THM4 levels measured in the blood samples before showering ranged from 0.014 to >0.507 μg/L, with a median value of 0.044 μg/L (see Table 2). These THM4 concentrations were significantly lower than the THM4 concentrations measured in the blood of Cobb County participants before showering ($p < 0.01$). An analysis of blood results obtained from Corpus Christi participants after showering indicated that median THM4 levels increased as a result of that water-use activity ($p < 0.0001$). The range of THM4 levels measured in the blood after showering was 0.083−>0.658 μg/L, with a median value of 0.172 μg/L. This represents an approximately 4-fold increase in the median THM4 concentration in the blood as a result of showering.

The brominated THMs represented the majority of the THM4 in Corpus Christi tap water. Chloroform, bromodichloromethane, and dibromochloromethane each accounted for slightly more than one-fourth of the total THM4 concentration on a molar basis (see Table 3). Bromoform accounted for approximately 15% of the THM4 concentration. The distribution of THM species measured in the blood of Corpus Christi participants was significantly different than the distribution measured in Cobb County participants due to the higher proportion of the brominated compounds ($p < 0.0001$).

Figures 1−3 show frequency distributions for the individual THM species in Corpus Christi tap water and in BS and AS blood samples. (Similar figures are not shown for Cobb County because chloroform dominated all three distributions.) While the order of occurrence of the species is the same for all sets of samples, striking differences in the proportions of chloroform were observed among the blood and water samples in Corpus Christi. Both sets of blood samples in Corpus Christi had significantly higher mole fractions of chloroform than the corresponding tap water samples ($p = 0.0001$). The median mole fraction of chloroform measured in the tap water was 0.286 (see Table 3). The blood analysis before showering showed a much higher median mole fraction of 0.680. Showering drove the median mole fraction of chloroform in the blood down to 0.427. This resulted in a distribution of THM species in the blood after showering that was more consistent with the distribution found in tap water.

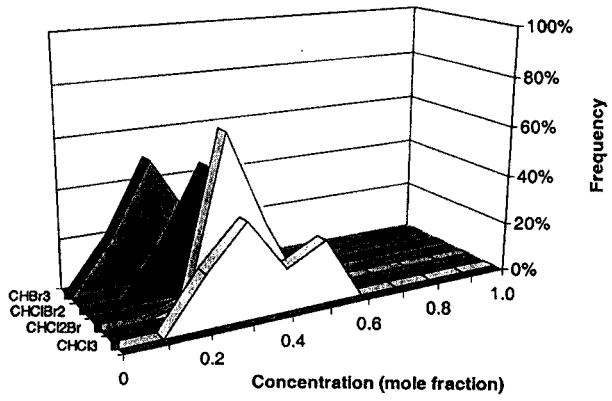

FIGURE 1. Frequency distributions for individual THM species measured in Corpus Christi tap water samples ($n = 24$).

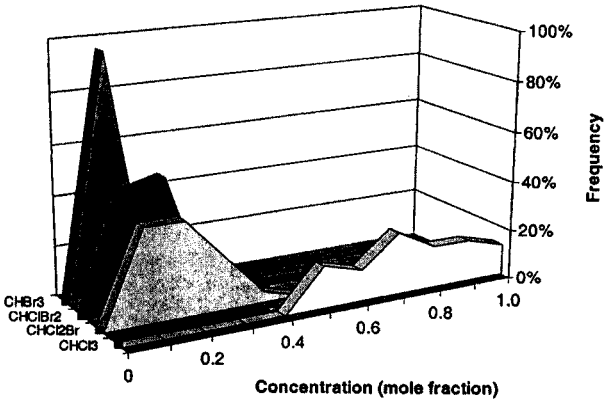

FIGURE 2. Frequency distributions for individual THM species measured in Corpus Christi blood samples before showering ($n = 24$).

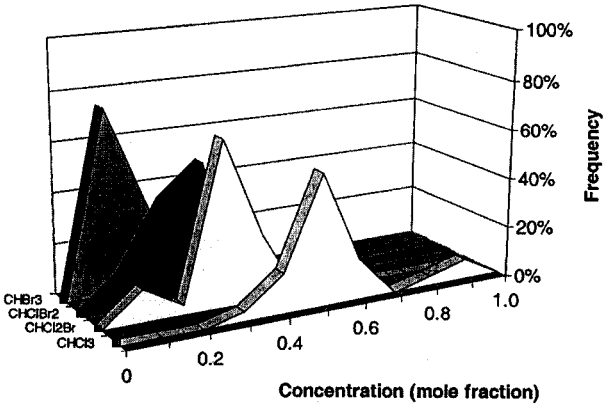

FIGURE 3. Frequency distributions for individual THM species measured in Corpus Christi blood samples after showering ($n = 24$).

The median BIF for Corpus Christi was higher than that of Cobb County, reflecting the higher concentration of bromide in the Corpus Christi water and thus the tendency for this water to favor formation of the brominated THM species. The median BIF values for tap water and BS and AS blood samples in Corpus Christi were 1.285, 0.549, and 0.910, respectively (see Table 3). These results indicate that the THM species measured in the blood of Corpus Christi residents tended to include less of the brominated species than the THMs measured in the participants' tap water samples. While showering increased the concentration of each THM species in the blood, it tended to increase the proportion of the brominated species, driving the distribution to be more consistent with that in the tap water ($p = 0.0001$).

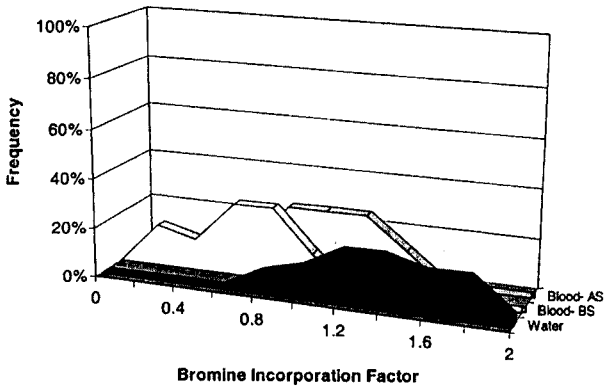

FIGURE 4. Frequency distributions for bromine incorporation factor (BIF) for Corpus Christi tap water and BS and AS blood samples ($n = 24$).

TABLE 4. Spearman's Correlation Coefficients (r_s) for Tap Water Concentrations and Before Showering (BS) or After Showering (AS) Blood Concentrations

blood–water correlation (r_s)		Cobb County, GA ($n = 24$)	Corpus Christi, TX ($n = 24$)
$CHCl_3$	BS	0.302	0.109
	AS	0.073	0.387[a]
$CHCl_2Br$	BS	0.157	0.337[a]
	AS	0.093	0.242
$CHClBr_2$	BS	0.070	−0.179
	AS	0.229	−0.043
$CHBr_3$	BS	nd[b]	0.050
	AS	nd	0.450[c]
THM4	BS	0.341[a]	0.035
	AS	0.138	0.068

[a] $p < 0.10$. [b] nd, not determined because bromoform was not detected in Cobb County tap water. [c] $p < 0.05$.

Figure 4 shows the BIF frequency distributions for Corpus Christi's tap water and blood samples.

Blood–Water Correlations. Figures 5 and 6 show sample plots of chloroform and bromodichloromethane concentrations measured in the blood before showering versus tap water concentrations. These graphs illustrate a general trend that as the THM concentrations in tap water increased, the corresponding THM concentrations in blood also tended to increase, although the data do not reflect a simple linear relationship. Table 4 lists the Spearman's correlation coefficients (r_s) for tap water and BS or AS blood results, with footnotes to denote those correlation coefficients that were statistically significant up to the 10% level.

In Cobb County, the correlation between tap water THM4 and BS blood THM4 concentrations was statistically significant with a correlation coefficient of 0.341 ($p = 0.10$). In Corpus Christi, statistically significant correlations were found between tap water and blood for chloroform AS ($r_s = 0.387$, $p < 0.10$), bromodichloromethane BS ($r_s = 0.337$, $p = 0.10$), and bromoform AS ($r_s = 0.450$, $p < 0.05$).

Discussion

This is the first study to evaluate the relationship between THM concentrations in the blood and THM concentrations in tap water. THM concentrations in the blood were measured to be 3 orders of magnitude lower than those in the tap water. The distribution of THM species in the subjects' blood samples in Corpus Christi was distinctly different from the distribution in Cobb County subjects, reflecting the differences in brominated species in Corpus Christi tap water as compared to Cobb County tap water. The inhalation and dermal exposure to THMs from showering resulted in an

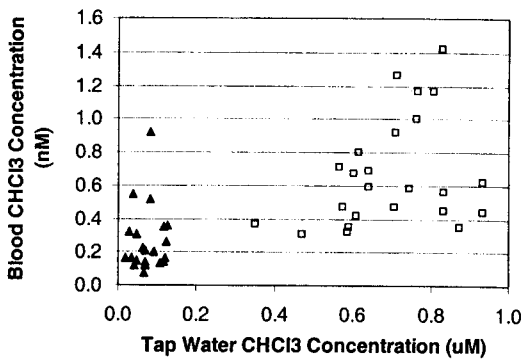

FIGURE 5. Relationship between molar $CHCl_3$ concentrations in blood before-showering and corresponding tap water for both sampling locations ($n = 48$): □, Cobb County data; ▲, Corpus Christi data. Note: Two outliers from Corpus Christi data are excluded from this graph but included in the analysis.

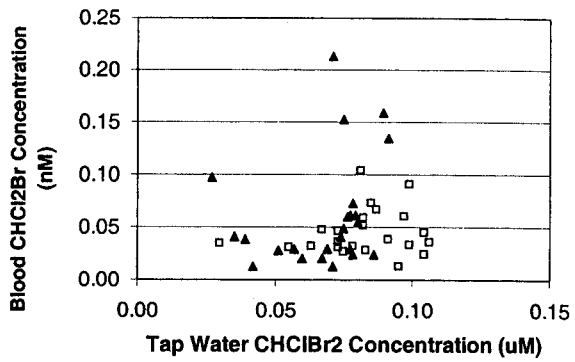

FIGURE 6. Relationship between before-showering blood $CHCl_2Br$ molar concentrations and corresponding tap water $CHCl_2Br$ molar concentrations for both sampling locations ($n = 48$): □, Cobb County data; ▲, Corpus Christi data.

approximately 4-fold increase in the median concentrations of THMs in the blood. In Corpus Christi, a much higher proportion of chloroform was found in the subjects' blood prior to showering relative to that present in the tap water. However, exposure to THMs from showering shifted the THM distribution to be closer to the distribution found in tap water. A simple linear relationship between tap water and blood THM concentrations was not observed.

There are a number of factors that may have contributed to the variations observed in blood THM concentrations that could not be explained by the tap water THM concentrations. In this study, the tap water concentrations were characterized by analyzing samples collected from a one-time sampling event in the participants' homes. The samples collected in this study, therefore, only represent a "snapshot" of tap water concentrations and may have been insufficient to capture the variability in tap water concentrations that affect blood THM concentrations. Significant temporal variability at the point of entry to the distribution system has been observed for both Cobb County and Corpus Christi (*27*). Also, tap water samples were not collected at the temperature used for showering. THM concentrations may increase when water is heated, depending on the chlorine residual of the water, and upon heating, the production of chloroform may be favored over the formation of the brominated species depending on the bromide ion concentration of the water (*28*). Therefore, it is possible that THM concentrations in the shower water were not consistent with those measured at the kitchen tap. Additionally, it took a finite amount of time to collect the blood samples after showering, during which time the THM species may have decreased as a result of metabolic processes (see below). The mean length of time between the end of the shower and drawing of the blood

sample was 11.7 min (standard deviation of 5.5 min), based on the 37 subjects for which we had valid times for both events. The median was 10.3 min with a range from 3.2 to 30.1 min.

The higher proportion of chloroform measured in the blood as compared to the tap water of both study groups, but particularly in Corpus Christi, could be associated with (i) the time delay in taking the blood samples after showering; (ii) different metabolic behaviors of the individual THM species; (iii) contributions made by ambient indoor air exposure; or (iv) additional sources of THM exposure. Parameters such as absorption kinetics and efficiency, excretion rates, metabolic rates, and the presence of genetic polymorphisms will affect THM concentrations and speciation in the blood. Pharmacokinetic experiments indicate that the brominated species are metabolized more quickly than chloroform, potentially resulting in a higher proportion of chloroform in the blood several hours after the exposure (*15*, *29*). This would be most pronounced in the BS blood samples, as there were presumably no major tap water exposures during the night. This could be the principal reason the bromine incorporation factor in the BS blood samples was low as compared to the corresponding tap water concentrations. Likewise, because the AS blood samples were delayed for up to 30 min after showering, there may have been a shift in the distribution of THM species during that time. As brominated species are metabolized more quickly than chloroform, the concentrations of the brominated species may have been higher at the end of the shower than the concentrations measured. Despite this, the results show that the THM distribution in the blood as a result of showering did shift toward the distribution of THMs measured in tap water.

It has been suggested that differences in metabolic rates among the THM species may reflect a genetic polymorphism (*17*, *29*). This may be a possible explanation for the apparent clustering of chloroform concentrations into a lower range (blood concentrations from 0.3 to 0.6 nM for tap water concentrations of 0.35–0.95 μM) and a higher range group (blood levels from 0.3 to 1.4 nM for the same tap water concentrations) in Cobb County (see open squares in Figure 5).

Differences in water-use activities may manifest in differences in the distribution of THM species found in the ambient indoor air. Parallel data from the survey questionnaire (*20*) showed that the number and length of showers per week were significantly different between Corpus Christi and Cobb County participants. There were other differences in water-use activities, such as bathing of children and dishwashing by hand, but these were not significantly different (*20*). As chloroform is the most volatile of the THM species, the THM distribution in ambient indoor air may be different than that measured in the tap water, resulting in higher exposure to chloroform through inhalation, especially for Corpus Christi residents. The Henry's law constants for chloroform and bromoform vary by an order of magnitude ($1.8–5.3 \times 10^{-3}$ atm-m^3/mol for $CHCl_3$ and 5.35×10^{-4} atm-m^3/mol for $CHBr_3$) (*30*, *31*). This would result in higher levels of chloroform in the blood than expected based on tap water concentrations alone.

Additional sources of exposure to THMs, particularly chloroform, may exist (*32*). For example, additional chloroform sources may include swimming pools, humidifiers, hot tubs, outdoor misters, and foods. This study did not address this issue.

Future work needs to focus on (i) obtaining tap water samples throughout the day to characterize the variability in tap water THM concentrations; (ii) measuring THM concentrations in tap water at the temperature of the exposure; (iii) identifying the rate and extent of THM volatilization and characterizing the distribution and variability of THM species in indoor air; (iv) evaluating the importance of sources of THMs beyond tap water; (v) developing an exposure model that includes specific water-use activities, duration of exposure, and each route of THM exposure; (vi) obtaining blood THM concentrations immediately after the exposure activity; (vii) evaluating the pharmacokinetic and physical–chemical parameters that influence the body burden in humans; and (viii) evaluating participants for a possible genetic polymorphism or other metabolic factors that influence the concentration of THMs in blood. The investigators are planning to conduct a more systematic study of these factors in future work.

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